

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

IN RE PCT NATIONAL STAGE APPLICATION OF Group Art Unit: **1796**
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INTERNATIONAL APPLICATION NO. **Confirmation No. 7040**
PCT/EP2005/051133
FILED: **March 14, 2005**
FOR: FUNGICIDAL DETERGENT
 COMPOSITIONS
U.S. APPLICATION NO: **10/593,227**
35 USC 371 DATE: September 18, 2006

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER RULE 132

I, Dietmar Ochs, a citizen of the Federal Republic of Germany, presently residing in 79650 Schopfheim, Germany, hereby declare:

CREDENTIALS

1. That I was awarded the degree of a Doctor of Natural Science by the University of Tübingen, Germany in 1990;

2. That I have been employed by CIBA Grenzach GmbH (now part of BASF SE) as research chemist in 1995;
3. That I hold presently a position as Head of Development/Technical Service Biocides at Ciba Grenzach, GmbH (now part of BASF SE);
4. That I have been engaged in R&D in the field of biocides in between 1995 and now;
5. That the experiments described in the following have been carried out under my supervision:

COMPARATIVE PROCEDURES

Composition of AATCC Liquid Detergent

LAS	12.1%
Linear Alcohol Ethoxylate	8%
Propane Diol	8%
Citric Acid	1.2%
Fatty Acid Ethoxylate	4%
NaOH	4%
Water	Balance

Tested microorganisms: Escherichia coli ATCC 10536

Chaetomium globosum ATCC 6205

Washing conditions (common standard Lini-Test conditions)

Detergent:	25 gram liquid detergent / Kg fabric
Liquor ratio:	1:5
Fabric:	20 g Cotton
Washing time:	20 minutes
Washing cycle:	1
Rinsing:	once during 30 seconds under running drinking water

Drying: at 50-60°C

Contact time: Immediately after contamination, after 6 and 24 hours at 37°C (bacteria)
 Immediately after contamination and after 1 week at 29°C (fungi)

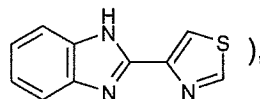
Nutrient medium: Caso agar (CA) with neutralizer (bacteria and yeast)
 Sabouraud agar (fungi)

Neutralizer: Phosphate buffer containing 1% Tween 80 + 0.3% Lecithin

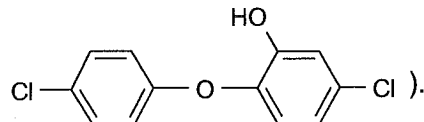
Diluent: Sterile deion. water

Incubation of plates: 24 hrs at 37°C (bacteria)
 4 days at 29°C (fungi)

Additives: Tinosan® Plus FG (active ingredient:



Tinosan® HP 100 (active ingredient:



Principle of Testing

Washed swatches with a diameter of 4 cm were inoculated with 500 µl of the bacterial or fungal suspension (= about $\sim 10^5$ cfu / sample), placed in a humid chamber and incubated at 37°C (bacteria) or 29°C (fungi). Each sample and contact time was tested twice.

Immediately after inoculation, after 6 and 24 hours at 37°C (bacteria) and immediately after contamination and after 1 week at 29°C (fungi), the samples were given in a sterile bag (Stomacher bag 80) containing 10 ml phosphate buffer 0.07 molar, pH 7.4 containing 1% Tween 80 and 0.3% lecithin and treated in the Stomacher for 1 minute. After shaking 1:10 dilutions until 10^{-4} in sterile deionised water were made.

From the undiluted and from the dilutions, samples of 100µl were plated out by the mean of a spiral plater. After incubation the surviving colonies were counted and reported in a table as cfu / sample.

Results (cfu / sample / log reductions)

Test strain -->	Escherichia coli ATCC 10536			
	0'	After 6 hrs	After 24 hrs.	Log red after 24 hrs *
AATCC Standard liquid detergent containing 0.2% TINOSAN® Plus FG	2.9 x 10 ⁵ 2.7 x 10 ⁵	> 10 ⁶ > 10 ⁶	> 10 ⁶ > 10 ⁶	< 1
AATCC Standard liquid detergent containing 0.6% TINOSAN® HP 100	2.9 x 10 ⁵ 2.7 x 10 ⁵	1.0 x 10 ³ 9.1 x 10 ²	< 100 < 100	> 5
AATCC Standard liquid detergent containing 0.1% TINOSAN® Plus FG + 0.3% TINOSAN® HP 100	2.9 x 10 ⁵ 2.7 x 10 ⁵	2.1 x 10 ³ 1.7 x 10 ³	4.6 x 10 ³ 3.8 x 10 ³	4.3
AATCC Standard liquid detergent without active (placebo)	2.9 x 10 ⁵ 2.7 x 10 ⁵	1.5 x 10 ⁷ 1.4 x 10 ⁷	6.9 x 10 ⁷ 8.2 x 10 ⁷	---

* calculated versus placebo (sample 4) after 24 hours at 37°C

O/n culture: Escherichia coli ATCC 10536

2.6 x 10⁹ / ml 1:1000 0.5ml / 2 discs

Test strain -->	Chaetomium globosum ATCC 6205		
	0'	After 1 week	Log red after 1 week *
AATCC Standard liquid detergent containing 0.2% TINOSAN® Plus FG	2.0 x 10 ⁷ 1.7 x 10 ⁵	5.1 x 10 ⁴ 6.6 x 10 ⁴	1.4
AATCC Standard liquid detergent containing 0.6% TINOSAN® HP 100	2.0 x 10 ⁷ 1.7 x 10 ⁵	4.2 x 10 ⁵ 2.3 x 10 ⁵	< 1
AATCC Standard liquid detergent containing 0.1% TINOSAN® Plus FG + 0.3% TINOSAN® HP 100	2.0 x 10 ⁷ 1.7 x 10 ⁵	1.0 x 10 ⁵ 1.1 x 10 ⁵	1.1
AATCC Standard liquid detergent without active (placebo)	2.0 x 10 ⁷ 1.7 x 10 ⁵	1.4 x 10 ⁶ 1.3 x 10 ⁶	---

* calculated versus placebo (sample 4) after 1 week at 29°C

Chaetomium globosum ATCC 6205 6.0 x 10⁶ / ml

1: 10

0.5ml / 2 discs

CONCLUSIONS

6. These results clearly indicate the broad spectrum of antimicrobial activity, in particular a simultaneous high activity against bacteria and fungi of a combination of 0.1% TINOSAN® Plus FG and 0.3% TINOSAN® HP 100 in a liquid detergent even at lower biocide concentration as compared to the individual ones.

7. These results are important because the instantly claimed method allows formulating a detergent for use in a domestic washing process which does not only clean the textile fiber material but also prevents or reduces affection with bacteria and fungi.

FINAL STATEMENT

I, Dietmar Ochs, declare further that all statements made herein of personal knowledge are true and all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Signed this 30 day of Oct. , 2009.



Dietmar Ochs